Composition of Soybean Lecithin

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ABSTRACT

Commercial soybean lecithin is a complex mixture containing ca. 65-75% phospholipids together with triglycerides and smaller amounts of other substances. The major phospholipids include phosphatidylcholine, phosphatidylethanolamine and inositol-containing phosphatides. Other substances reported include carbohydrates, pigments, sterols and sterol glycosides. This paper reviews the nature of the compounds found in soybean lecithin and our present knowledge of its composition.

INTRODUCTION

The term "lecithin" as used today refers to the material obtained by degumming crude vegetable oils and drying the hydrated gums. In the U.S., commercial lecithin is predominantly from soybean oil. The specific phospholipid formerly called lecithin is now referred to as phosphatidylcholine.

Lecithin, then, consists not only of a mixture of phospholipids but also of triglycerides and other nonphospholipid compounds removed from the oil in the degumming process. Composition of soybean lecithin has been considered in many reviews, and substances commonly reported included triglycerides, fatty acids, pigments, sterols, sterol glycosides and esters, tocopherols, and carbohydrates (1-6). Ranges in composition from some of these reviews are in Table I, and values for some other minor components (7) are in Table II. Composition of commercial lecithin is further complicated by the production of six grades of plastic and fluidized lecithin in unbleached, single-bleached and double-bleached forms (2,4,6). In addition, various other refined and modified materials are produced (5,6,8). Little has been published about the chemical nature of changes produced by peroxide bleaching or other modifications. This paper reviews the composition of the phospholipids and other materials extracted with the oil and removed by degumming as determined by studies of commercial and laboratory prepared materials. Wittcoff has discussed sovbean phosphatides in his comprehensive book "The Phosphatides" (9).

NONPHOSPHOLIPID COMPONENTS

Carr (10) has stated that oil content may be varied by changing the conditions of centrifugation, and ca. 7% soybean oil and 3% fatty acids are blended with the lecithin to increase fluidity.

Color of soybean lecithin depends on processing and bleaching conditions. At our laboratory (11), we showed that xanthophylls are preferentially removed with the gums and that carotene remains with the oil. Lutein made up about 75% of the carotenoid pigments in the gums. These carotenoid pigments are largely destroyed by peroxide bleaching, leaving a variable amount of brown color with no characteristic absorption bands. Studies by Zuev et al. (12-14) have confirmed the formation of brown-colored substances as well as destruction of carotenoids by heating. They suggest oxidation is involved. Although Burkhardt (15) found in safflower oil that phosphatidylethanolamine contributed most to color formation, Tomioka and Kaneda (16,17) found the brown products similar to those formed in an aldol condensation reaction rather than in a Maillard reaction.

Sterols and sterol glycosides have long been known in

soybean lecithin. In addition, Lepage (18) has isolated an esterified form, or acylated steryl glucoside. In a sample of crude soybean lecithin, Kiribuchi et al. (19) reported free sterols 0.5%, steryl glucoside 2.1% and acylated steryl glucoside 2.6%. Steryl ester was present in negligible amounts. In all three forms, campesterol, stigmasterol and β -sitosterol were present in a ratio of ca. 20:30:50. All fatty acids normally in soybean oil were found in the acylated steryl glucosides with increased amounts of saturated acids. Popov et al. (20) found 0.74% free sterol and 2.68% combined steryl glycoside and esterified steryl glycoside.

Although tocopherols are present in soybean lecithin, no recent values were found for their amount. Review articles (2,4,7) list a concentration of ca. 0.1%. One might expect that the proportions of individual tocopherols would be similar to soybean oil; a recent study of soybean oil (21) shows a ratio of α : γ : σ tocopherol of ca. 5:68:27.

These oil-soluble nonphosphatide materials are largely separated from the phospholipids by extracting with acetone. Some sterol glycosides and free carbohydrates, as well as sugars bound to lipid constituents, remain with the acetone-insoluble phospholipids. At our laboratory (22), one sample of oil-free phosphatides was found to contain 7% free sugars made up of 45% sucrose, 9% raffinose and 47% stachyose.

PHOSPHOLIPID COMPONENTS

Each of the phospholipid classes is a mixture of any individual compounds with different fatty acids. Reported fatty acid compositions of total acetone-insoluble phosphatides vary considerably. Some values are listed in Table III. Although the fatty acids of phosphatide classes differ, little has been published on pure materials isolated so as to preclude fractionation. Another complexity is added to

TABLE I

Reported Range of Components of Soybean Lecithin (1-6)

	%
Phosphatidylcholine	19-21
Phosphatidylethanolamine	8-20
Inositol phosphatides	20-21
Other phosphatides	5-11
Soybean oil	33-35
Sterols	2-5
Carbohydrates, free	5
Moisture	1

TABLE II

Some Minor Components of Soybean Lecithin (7)

Tocopherol Biotin Folic acid Thiamin Riboflavin Panothenic acid	1.3 mg/g 0.42 µg/g 0.60 µg/g 0.115 µg/g 0.33 µg/g 5.59 µg/g

TABLE III

Some Reported Fatty Acid Compositions of Soybean Phosphatides

	Hilditch and Zaky (23) (%)	Vijayalakshime and Rao (24) (%)	Daga (25) (%)
14:0		-	1.9
14:1	****	·	tr
16:0	11.7	42.7	26.7
16:1	8.6	7.0	-
18:0	4.0	11.7	9.3
18:1	9,8	17.0	25.1
18:2	55.0	20.0	27.0
18:3	4.0	1.6	_
20:0	1.4		
Unsaturated C ₂₀₋₂₂	5.5	_	_

phosphatide composition by the different cations associated with acidic groups. Potassium, sodium, magnesium and calcium have been reported (2).

Of the phospholipids in soybean lecithin, the best known is phosphatidylcholine (PC), for which the structure is shown in Figure 1. This formula represents the α isomer. Hydrolysis of PC results in both α and β glycerophosphoric acid, and this was formerly considered evidence for both α and β PC. However, Baer (26) has shown that hydrolysis of the synthetic compounds L-\alpha-glycerylphosphorylcholine, L-α-glycerylphosphorylethanolamine, L-α-PC, or L-α-phosphatidylethanolamine (PE) all yield mixtures of α - and $\hat{\beta}$ -glycerophosphoric acid that resemble closely those reported from natural PC. He states that the concept of the occurrence of β -PC or β -PE in nature is no longer tenable. Privett and Blank (27) and Mangold (28), by ozonolysis of PC and thin layer chromatography of the resulting aldehyde "core," have shown four fatty acid groups in PC: α-saturated- β -unsaturated, α -unsaturated, β -saturated, α - β -unsaturated, and a small amount of α - β -saturated. Kimura et al. (29) by AgNO₃-TLC separated soybean PC into four fractions, including one containing 64% arachidonic acid. Recently, Crawford et al. (30) by high pressure reverse-phase chromatography have isolated fractions from soybean PC ranging in unsaturation from dilinolenyl to stearyl, linolyl PC.

In older literature, the term "cephalin" has been used for PE (Fig. 1) and as "cephalin fraction" for material insoluble in alcohol. Actually, soybean PE has been found both in the alcohol and alcohol insoluble fractions (31-33), and a 30 transfer countercurrent distribution of the alcohol-soluble fraction between hexane and 90% methanol resulted in only partial separation of PE from the completely alcohol-soluble PC. Kimura et al. (34), by methods like they used with PC, also found arachidonic acid in soybean PE.

Closely related to PE is phosphatidylserine (PS), found in soybean lecithin by Van Handel (35). Negishi et al. (36) reported 5.9% PS in a commercial soybean lecithin and Nielson (37) found PS in difficultly extractable soybean phosphatides.

The structure of N-acylphosphatidylethanolamine (NPE) in soybean lecithin was determined by Aneja et al. (38). Wilson and Rinne (39) found it to be more abundant in developing seed and to decrease at maturity.

Phosphatidic acid (PA), the moiety common to all the glycerophosphatides, is present in soybean lecithin (1,2,5). It, also, has been found in greater amounts of lipids from developing than from mature seeds (39, 40). Both PA and NPE are decreased by extraction procedures that inactivate enzymes (41, 42). The amount of NPE and PA in lecithin, therefore, may be dependent on enzymatic reactions during storage and processing of the beans, and PA as calcium and

magnesium salts also remains in the degummed oil as nonhydratable phosphatides (43, 44).

Inositol was first found in soybean phosphatides by Klenk and Sakai (45), and early work was done by Woolley (46) and Folch (47). At our laboratory, the inositol lipids in soybean phosphatides were separated into two fractions by countercurrent distribution between hexane and 95% methanol (31). The fraction of lower partition coefficient—more soluble in methanol—was found to contain PE and an inositol-containing phosphatidic acid (32). This inositol compound corresponds to phosphatidylinositol (PI) for which the structure was determined by Okuhara and Nakayama (48). Its absolute configuration was later shown to be as in Figure 1 (49). The fraction of higher partition coefficient—more soluble in hexane—contained, in addition to inositol, the sugars galactose, mannose and arabinose (22).

These complex glycolipids and similar materials from other sources have been very difficult to purify and characterize. They were the object of much work by Carter and his group, and a list of his papers on this and related subjects has been published (50). Van Handel (35, 51) found evidence for a long chain base in soybean phosphatides, and Carter et al. (52,53) identified phytosphingosine and dehydrophytosphingosine (1,3,4-trihydroxy-2-amino-octadecane and 1,3,4,-trihydroxy-2-amino-8-trans-octadecene) in soybean phosphatides in the ratio saturated:dehydro of 20:80.

FIG. 1. Some phosphatides in soybean lecithin.

FIG. 2. Phytoglycolipid tetrasaccharide from phospholipids.

In a series of papers (54-59), the complete structure of a phytoglycolipid (PGL) tetrasaccharide containing phytosphingosine, fatty acid, phosphorus, inositol, mannose, hexuronic acid and glucosamine was determined to be as shown in Figure 2. To this structure, arabinose, galactose and a small amount of fucose is attached through glucosamine to give various chain lengths, including octa- and higher oligosaccharides (57,58). Soybean PGL may be presumed to be mainly the unsaturated analog of the structure just discussed (53). In soybean PGL, the fatty acid composition was cerebronic acid 5% and palmitic plus stearic acids 95%.

Isolation of PGL involved alkaline hydrolysis of phospholipids and did not preclude possible cleavage of a more polar moiety (54). Purification by countercurrent distribution was unsatisfactory because impurities were bound as mixed chelated calcium and magnesium salts. This difficulty was overcome by passage of the lipids through a chelating resin column (60). Using countercurrent distribution of soybean phospholipids in the sodium form, Carter and Kisic (61) confirmed the presence of PGL as previously isolated by alkaline hydrolysis, and also found a second glycolipid, called ceramide-phosphate polysaccharide, devoid of hexosamine but containing inositol, hexuronic acid, galactose, arabinose, fucose and mannose. They also found a material, lipophytin, high in phosphorus and insoluble in their hexane, butanol, methanol, water solvent system. These last two substances still do not appear to have been further characterized.

Modern chromatographic methods have made possible the detection of many other materials in soybean lecithin. Although the major phospholipids are PC, PE and PI, many other components are present in small amounts. Szuhaj et al. (62), by silicic acid column and thin layer chromatography, found 17 classes as follows: triglycerides, diglycerides, monoglycerides, free fatty acids, free sterols, steryl esters, diphosphatidylglycerol, PE, PC, PI, lyso-PC, esterified steryl glucoside, steryl glucoside, digalactosyldiglycerides, cerebrosides and two unidentified glycolipids. Erdahl et al. (63) found most of the above and also NPE, lyso-PE, PS, phosphatidylglycerol and eight unidentified components.

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